

COMPARATIVE THERAPEUTIC EFFICACY OF HYPERTONIC SALINE SOLUTION AND ISOTONIC NORMAL SALINE FOR FORE-STOMACH IMPACTION DISORDERS IN BUFFALOES (*Bubalus bubalis*)

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ABSTRACT

The aim of present investigation was to evaluate the comparative therapeutic efficacy of isotonic (0.9%) saline solution (NSS) and hypertonic (7.5%) saline solution (HSS) along with supportive medical treatment in buffaloes suffered with forestomach impaction. A total of 26 buffaloes were undertaken in two groups. In Group 1, twenty buffaloes were infused with isotonic (0.9%) saline solution 20 ml/kg body weight and in Group 2 six buffaloes were infused with hypertonic saline solution (7.5%) 2 ml/kg. The physical parameters (heart rate and respiration rate) were decreased more significantly in Group 1 after treatment. Similar trend was observed in the pretreatment hematological parameters *viz.* Hb, PCV, TLC, neutrophil count and ESR after fluid therapy which was indicative of expansion of plasma volume. There was not much effect on rumen liquor parameters after fluid therapy. The biochemical profile (Aspartate aminotransferase (AST), Alkaline phosphatase (ALP), bilirubin, Blood Urea Nitrogen (BUN), creatinine, total protein, glucose, globulin and fibrinogen) was more significantly declined in Group 1 after fluid therapy, whereas plasma sodium and chloride

levels significantly increased, and plasma potassium level more significantly decreased in Group 2. The recovery rates were 60% and 17% respectively in Group 1 and Group 2. Thus, Isotonic (0.9%) saline solution appears to more effective in therapeutic management of forestomach impaction in buffaloes.

Keywords: *Bubalus bubalis*, buffaloes, hemato-biochemical, hypertonic saline solution, impaction, normal saline solution

INTRODUCTION

Impaction in dairy animals refers to the failure of digestion, slow passage, and accumulation of feed in one or more of the stomach compartments leading to constipation or absence of dung (Sharma *et al.*, 2015). The complex pathophysiological changes of forestomach impaction results in hypovolemia, disturbed electrolyte balance and consequent changes in acid base balance (Garry, 2002). As forestomach disorder results in moderate to severe alkalosis, non-alkalinizing solutions are fluid of choice for most dehydrated mature ruminants (Roussel, 1989).

Isotonic saline solution is used for acute plasma volume expansion, correction of hypovolemia and treatment of metabolic alkalosis (Ward *et al.*, 1993). Another alternative is hypertonic saline solution, which along with other symptomatic treatment offers a quicker, practical, economical, and most importantly an effective way for the treatment of mild to moderate cases of forestomach impaction in buffaloes (Saxena *et al.*, 2007). Hypertonic solutions promote immediate blood volume expansion, restore cardiac output and regional blood flows, and improve microcirculation (Polido-Figueiredo, 2006).

The incidence of fore-stomach impaction has increased enormously in past few years in Punjab state of India (Sharma *et al.*, 2015). The routine treatment given to such patient is often ineffective and they generally require rumenotomy to relieve impaction. There is a firm experimental basis for the actions of hypertonic saline solutions in hemorrhagic hypotension. These actions are presumed to be of benefit in the treatment of sepsis or septic shock. However, there is lack of data regarding the use of these solutions in clinical studies of large animals. So, keeping in view this fact, the present investigation deals with therapeutic efficacy of isotonic saline (0.9%) solution and hypertonic saline (7.5%) solution along with supportive treatment in forestomach impaction in buffaloes.

MATERIALS AND METHODS

The study protocol was performed in compliance with institutional guidelines. All owners gave consent for buffaloes to be included in the study. The study was conducted on 26 buffaloes presented at Large Animal Unit of

Teaching Veterinary Hospital, Guru Angad Dev Veterinary and Animals Science University (GADVASU), Ludhiana. The cases were taken at random on the basis of history of constipated feces or loss of defecation. All the buffaloes were examined clinically according to the standard procedures (Radostits *et al.*, 2000). On the basis of detailed clinical examination, a tentative diagnosis of forestomach impaction was established, and the animals were subjected to treatment. A total of twenty-six buffaloes were divided into following two groups:

Group 1

In this group twenty buffaloes were given isotonic saline (0.9%) solution, intravenously 20 ml/kg b.wt. along with supportive medicinal therapy.

Group 2

In this group six buffaloes were given hypertonic saline (7.5%) solution, intravenously 2 ml/kg b.wt. over a period of 2 hours along with supportive medicinal therapy.

The rate of fluid infusion was calculated as follows:

$$\text{Infusion rate} = \frac{\text{ml/kg b.wt.} \times \text{b.wt. (Kg)} \times \text{drops/ml}}{\text{Total time in min. drop/ml}}$$

Supportive treatment included i.v. injection of 450 ml of calcium-magnesium-borogluconate (8.37 g of calcium, 5.31 g phosphate, 2.08 g magnesium and 90 g of dextrose), amoxicillin and cloxacillin 15 to 20 mg/kg im (as per need), metoclopramide 0.2 mg/kg iv and herbal rumenototics orally and B complex 10 ml im and linseed oil (1 litre) was given orally as laxative.

Haematology

For determination of hematological

parameters, blood samples were collected aseptically from jugular vein in vials containing disodium ethylene diamine acetate (Na_2 EDTA) 2 mg/ml. Within 6 h of collection, the whole blood was used for estimation of Hemoglobin (Hb), Packed Cell Volume (PCV), Total Leucocyte Count (TLC), Differential Leucocyte Count (DLC) and Erythrocyte Sedimentation Rate by standard methods (Jain, 1986).

Clinical biochemistry

For determination of Plasma biochemical profile, blood samples were collected in heparinised vial. Plasma was separated and Total plasma protein (TP), Plasma albumin, Plasma globulin, Plasma glucose, Blood Urea Nitrogen (BUN), Plasma creatinine, Plasma calcium, Plasma chloride, Total bilirubin, Plasma enzymes activities (IU/L) viz. aspartate aminotransferase (AST) and alkaline phosphatase (ALP) were estimated by using Bayer's Diagnostic Kit with the help of RA50 Autoanalyser. Plasma Inorganic phosphorus, sodium and potassium were estimated by standard methods. Fibrinogen level was estimated by heat precipitation method (Jain, 1986).

Rumen liquor analysis

Rumen liquor samples were collected from left paralumber fossa and subjected to physical and microscopic examination for colour, odour, consistency, pH, and protozoa motility (Garry, 2002). A part of sample was filtered through double layer of muslin cloth and centrifuged. The supernatant was divided into two parts, one was preserved with few drops of saturated mercuric chloride solution for total volatile fatty acid (TVFA) and total ammonia-nitrogen (NH_3 -N) estimation, while the other one for sodium and chloride estimation. Rumen Sodium and chloride,

Sedimentation activity time (SAT) Methylene Blue Reduction Test (MBRT), Total Volatile Fatty Acid (TVFA) and Total ammonia-nitrogen (NH_3 -N) were estimated by standard methods.

Statistical analysis

For statistical analysis of data students "t" test and ANOVA tests were performed. The significance was seen at $P < 0.05$ and $P < 0.01$ level (Snedecor and Cochran, 1989).

RESULTS AND DISCUSSIONS

In Group 1, all the 20 buffaloes were suspected for fore-stomach impaction were treated with isotonic (0.9%) normal saline solution with supportive medicinal therapy, twelve animals showed clinical recovery within 24 to 72 h after treatment. The clinical recovery was manifested by improvement in clinical signs, passing feces and urine, rumination and normal intake of feed and water. Out of the 12 recovered animals, 5 had rumen impaction, 2 had mild peritonitis and 5 had impaction in late pregnancy. The rumen impaction was mild to moderate. The duration of disease in impaction in late pregnancy was 3 to 5 days, in peritonitis 7 to 10 days, in rumen and omasal impaction it was 5 to 15 days. The remaining eight animals with duration of disease for 7 to 20 days and did not recover with fluid and medicinal therapy were subjected to rumenotomy for evacuation of forestomach contents. Out of these eight animals, five had severe rumen impaction and three had severe omasal impaction. So, from 10 animals with rumen impaction, 5 with mild to moderate rumen impaction showed recovery while 5 with severe rumen impaction had to undergo rumenotomy.

In Group 2, 6 buffaloes were treated with

hypertonic (7.5%) saline solution with supportive medicinal therapy, but only one animal showed clinical recovery within 24 h of treatment while remaining five animals had shown improvement in clinical signs like passing urine and increased water intake but did not pass feces. The duration of disease in these cases varied from 10 to 20 days. Similar to the first group, severe rumen and omasal impaction cases did not respond to fluid therapy alone. Five non-recovered animals underwent rumenotomy for diagnosis of condition and evacuation of forestomach contents. Out of five, three had severe rumen impaction and two had severe omasal impaction. We found that fluid therapy, whether isotonic or hypertonic helps in mild to moderate rumen impaction while late pregnancy may need induction of parturition and severe impaction need surgical intervention.

The pretreatment physical parameters (heart rate, respiration rate) decreased significantly ($P \leq 0.05$) in both groups (Table 1). The decrease in respiration rate was more pronounced in Group 1 than Group 2 (Table 2). The decrease in heart rate and respiration rate may be due to increase in dynamic efficiency of the circulatory system (Velasco *et al.*, 1980). Similar results were reported by Saxena *et al.* (2007) but Ward *et al.* (1993) observed no significant differences between isotonic and hypertonic saline solution treatment with regard to vital signs and physiological variables like heart and respiration rate. Singh *et al.* (2007) reported decreased heart rate after hypertonic saline solution treatment in bovine with endotoxemic shock.

The pretreatment hematological parameters *viz.*, Hb, PCV, TLC and ESR decreased significantly ($P \leq 0.05$) in Group 1 after fluid therapy and non-significantly in Group 2 (Table 3 and 4). The decline in Hb level might be due to expansion

of plasma volume (Cambier *et al.*, 1997). The significant decrease in Hb level in Group 1 could be due to more amount of fluid infused. In cows with endotoxin induced mastitis, small volumes of isotonic or hypertonic saline solutions resulted in decrease in haematocrit due to increased plasma volume (Tyler *et al.*, 1994). After infusion of hypertonic saline in dogs, decreased PCV was observed by Constable *et al.* (1994). Similarly decreased haematocrit was reported after isotonic saline infusion in dogs with septic shock (Fantoni *et al.*, 1989). The mean relative neutrophil count in both groups showed decreasing trend after fluid therapy, but these variations were not significantly different from their respective pretreatment level. The mean lymphocyte, eosinophil, monocyte, and basophil counts did not show any significant change with respect to pretreatment level in both groups. The decrease in neutrophil count can be attributed to a decrease in circulating corticosteroids in the body due to decreased stress. The decrease in ESR can be explained by increased plasma volume (Tyler *et al.*, 1994), which corrects the dehydration and thus reduces plasma protein particularly fibrinogen and globulin, leading to decrease in ESR by altering the electrical charge on erythrocytes as reported by Jain (1986); Brar *et al.* (2002).

There were not much appreciable changes were observed in rumen liquor colour, consistency and pH but odour become less pungent following fluid therapy. Protozoa motility slightly improved from pre treatment pretreatment level, which was indicated by slight increase in TFVA. There was significant decline ($P < 0.05$) in $\text{NH}_3\text{-N}$ in Group 1 (Table 5 and 6). There were no appreciable changes in MBRT and SAT which indicated persistent microbial inactivity, because microflora require prolonged period to acclimatized in rumen environment (Garry, 1990). There was significant

($P < 0.05$) rise in sodium concentration in Group 2 which might be attributed to higher concentration of Na in infused fluid. There was no appreciable change in ruminal chloride content in both groups. No literature could be traced out for comparing effect of fluid therapy on rumen fluid variables.

The post treatment mean AST level decreased significantly ($P < 0.05$) in both groups but more pronounced in Group 1 than Group 2. The post treatment mean ALP, bilirubin, glucose levels in Group 1 was significantly lower ($P < 0.05$) than pretreatment level whereas in Group 2 declining trend did not differ significantly from pretreatment level (Table 7). It may be due rapid plasma volume expansion (Jean *et al.*, 1993). These changes were attributed to dilution of circulatory blood, reducing stress on vital organs induced by increased cardiac output and mean arterial pressure (Singh *et al.*, 2002). This may be due to resuscitative effects by rapid plasma volume expansion, osmotically drawing intracellular and interstitial water into the vascular space which is approx. 3 ml for every 1 ml of hypertonic saline solution infused (Smith *et al.*, 1985). The post treatment mean BUN and creatinine levels in both groups were significantly ($P < 0.05$) lower than their respective pretreatment value. The declining trend was more pronounced in Group 2 due to more infusion amount. The decrease in BUN may be attributed to increased renal flow following fluid therapy, explained by high cardiac output (Roch-e-Silva *et al.*, 1986; Kien *et al.*, 1991). Release of atrial natriuretic factor may also contribute to increasing urine output and thereby decreasing BUN level (Richards *et al.*, 1985). The mean total plasma protein, globulin and fibrinogen levels significantly decreased ($P < 0.05$) in Group 1 after fluid infusion, whereas in Group 2 plasma total protein and globulin showed declining trend but did not differ significantly from their respective

pretreatment level. There were no significant changes in plasma albumin concentration in both groups. Decrease in total protein and globulin may be attributed not only to dilution of circulatory blood but also to the osmotic effects of the solutions (Velasco *et al.*, 1980; Schertel, 1990; Constable *et al.*, 1991).

The post treatment mean plasma calcium levels in both groups were significantly higher ($P < 0.05$) than their respective pretreatment level. It may be due to infusion of calcium borogluconate as supportive therapy. The post treatment mean plasma phosphorus levels in both groups had decreasing trend but did not differ significantly from their respective pre treatmentpretreatment levels. The decrease in plasma phosphorus level may be due to increased GFR, which causes excretion of phosphorus (Roeder *et al.*, 1997). The plasma sodium and chloride concentration significantly increased ($P < 0.05$) and plasma potassium significantly ($P < 0.05$) decreased in both groups following fluid therapy but this tendency was more pronounced in Group 2 (Table 8). It may be attributed to the high osmolar sodium chloride load from the infusion (Moon *et al.*, 1991) and it indicated a rapid equilibrium with extra vascular fluid compartments (Tyler *et al.*, 1994). The resultant volume expansion is attributed to HSS osmotically drawing intracellular and interstitial water into vascular space (Smith *et al.*, 1985). The serum sodium and chloride concentrations, plasma and rhythm of glomerular filtration were restored after use of small volumes of hypertonic saline solution in a single dose. There was less marked hemodilution due to hypertonic saline and its administration brought serum potassium concentration and rhythm of glomerular filtration to normal level (Flores, 2006). Similar results were reported by Saxena (2003) in buffaloes with

Table 1. Effect of fluid therapy on physical parameters in buffaloes with forestomach impaction.

Parameters	Temperature (F°)	Heart rate (per min)	Respiration rate (per min)
Group 1 (Before) N=20	102.15±0.32	74.35±3.93	28.55±1.88
Group 1 (After) N=20	101.66±0.15	61.15±1.90*	19.25±0.94*
Group 2 (Before) N=6	102.60±0.30	69.50±3.36	18.33±2.36
Group 2 (After) N=6	101.80±0.16	58.16±2.83*	14.16±0.90

*Difference significant at P<0.05.

Table 2. Comparison of fluid therapy on physiological parameters in buffaloes with forestomach impaction.

Parameters	Temperature (F°)	Heart rate (per minute)	Respiration rate (per minute)
Group 1 (After) N=20	101.66±0.15	61.15±1.90	19.25±0.94*
Group 2 (After) N=6	101.80±0.16	58.16±2.83	14.16±0.90

*Difference significant at P<0.05.

Table 3. Effect of therapy on haematological parameters in buffaloes with forestomach impaction.

Parameters	Hb (gm%)	PCV (%)	TLC (x10 ³ µL ⁻¹)	N (%)	L (%)	E (%)	M (%)	B (%)	ESR (mm in one hour)
Group 1 (Before) N=20	7.80±0.40	40.60±1.73	10.00±1.0	67.95±2.14	29.80±2.06	1.15±0.30	0.60±0.18	0.05±0.05	98.75±6.11
Group 1 (After) N=20	7.00±0.35*	32.15±1.56*	6.80±0.54*	54.05±1.80	46.00±2.15	2.85±1.66	0.15±0.08	0.00±0.00	71.25±3.36*
Group 2 (Before) N=6	7.45±0.52	30.00±2.23	6.70±0.96	66.00±3.18	31.66±2.80	1.83±0.70	0.50±0.22	0.00±0.00	84.00±8.85
Group 2 (After) N=6	7.20±0.64	27.50±2.29	5.60±0.4	64.16±2.00	53.83±1.24	1.50±0.71	0.50±0.22	0.00±0.00	74.00±6.11

Table 4. Comparison of fluid therapy on haematological parameters in buffaloes with forestomach impaction.

Parameters	Hb (gm%)	PCV (%)	TLC (x10 ³ µL ⁻¹)	N (%)	L (%)	E (%)	M (%)	B (%)	ESR (mm in one hour)
Group 1 (After) N=20	7.00±0.35*	32.15±1.56*	6.80±0.54*	54.05±1.8	46.00±2.15	2.85±1.66	0.15±0.08	0.00±0.00	71.25±3.36*
Group 2 (After) N=6	7.20±0.64	27.50±2.29	5.60±0.4	64.16±2.00	53.83±1.24	1.50±0.71	0.50±0.22	0.00±0.00	74.00±6.11

*Difference significant at P≤0.05.

Table 5. Effect of fluid therapy on rumen liquor parameters in buffaloes with forestomach impaction.

Parameters	pH	MBRT (min)	TVFA (mEq/L)	NH ₃ N (mg%)	Na (mmol-l ⁻¹)	Cl ⁻ (mmol-l ⁻¹)
Group 1 (Before) N=20	7.20±0.13	45.35±1.70	63.30±2.23	37.05±2.13	118.70±3.50	23.84±1.70
Group 1 (After) N=20	7.00±0.20	38.00±1.28	77.70±2.30	27.95±1.00*	121.05±2.91	25.96±0.60
Group 2 (Before) N=6	6.66±0.21	43.66±3.43	75.66±9.10	27.50±1.89	113.83±5.78	23.25±1.62
Group 1 (After) N=6	7.00±0.00	40.21±3.00	82.33±7.98	23.16±0.87	125.2±1.36*	24.16±2.63

*Difference significant at P≤0.05.

Table 6. Comparative observations of fluid therapy on rumen liquor parameters in buffaloes with forestomach impaction.

Parameters	pH	MBRT (min)	TVFA (mEq/L)	NH ₃ N (mg%)	Na (mmol-l ⁻¹)	Cl ⁻ (mmol-l ⁻¹)
Group 1 (After) N=20	7.00±0.20	38.00±1.28	77.70±2.30	27.95±1.00*	121.05±2.91	25.96±0.60
Group 1 (After) N=6	7.00±0.00	40.21±3.00	82.33±7.98	23.16±0.87	125.20±1.36*	24.16±2.63

*Difference significant at P<0.05.

Table 7. Effect of fluid therapy on biochemical parameters in buffaloes with forestomach impaction.

Parameters	AST (IU/L)	ALP (IU/L)	Bilirubin (mg%)	Glucose (mg%)	BUN (mg%)	Creatinine (mg%)	TP (gm%)	ALB (gm%)	GLB (gm%)	Fibrinogen (mg%)	Ca (mg%)	P (mg%)	Na (m mol-l ⁻¹)	K (m mol-l ⁻¹)	Cl ⁻ (m mol-l ⁻¹)
Group 1 (n=20) (Before)	180.9±	181±	1.7±	98.5±	57.09±	1.74±	8.4±	3.35±	5.11±	980.50±	7.1±	6.95±	119±	3.14±	84.5±
Group 1 After	11.91	11.00	0.31	6.1	7.19	0.21	0.12	0.13	0.14	79.56	0.35	0.16	1.45	0.04	1.40
Group 2 (n=6) Before	127±	137.65±	0.9±	66.25±	35.95±	1.20±	7.76±	3.56±	4.28±	720.00±	9.24±	6.85±	135.45±	2.99±	96.26±
Group 2 After	10.30*	6.54*	0.18*	2.10*	3.90*	0.15*	0.13*	0.13	0.18*	65.53*	0.16	0.12	1.97*	0.17*	1.37
Group 2 N=6	182.16±	195.33±	1.5±	96.83±	68.23±	2.00±	8.2±	3.16±	5.28±	1033.33±	6.98±	7.0±	117±	3.04±	89.15±
Group 2 N=6	21.79	27.33	0.64	9.04	7.96	0.19	0.33	0.36	0.64	158.46	0.50	0.01	3.75	0.15	2.97
Group 2 N=6	150.16±	166.83±	1.0±	86.66±	57.61±	1.40±	7.8±	3.61±	4.90±	924.56	8.9±	6.90±	140.33±	2.77±	105.21±
Group 2 N=6	15.74	22.76	0.29	4.21	6.59*	0.09*	0.34	0.29	0.42	±40.96	0.10*	0.10	3.07	0.11*	2.09*

*Difference significant at P≤0.05.

Table 8. Comparison of fluid therapy on biochemical parameters in buffaloes with forestomach impaction.

Parameters	AST (IU/L)	AKP (IU/L)	Bilirubin (mg%)	Glucose (mg%)	BUN (mg%)	Creatinine (mg%)	TP (gm%)	ALB (gm%)	GLB (gm%)	Fibrinogen (mg%)	Ca (mg%)	P (mg%)	Na (m mol-l ⁻¹)	K (m mol-l ⁻¹)	Cl ⁻ (m mol-l ⁻¹)
Group 1 After N=20	127±	137.65±	0.9±	66.25±	35.95±	1.2±	7.76±	3.56±	4.28±	720.00±	9.24±	6.85±	135.45±	2.99±	96.26±
Group 2 After N=6	10.30*	6.54*	0.18*	2.10*	3.90*	0.15	0.13*	0.13	0.18*	65.53	0.16	0.12	1.97	0.17	1.37
Group 2 After N=6	150.16±	166.83±	1.0±	86.66±	57.61±	1.4±	7.8±	3.61±	4.90±	924.56±	8.9±	6.90±	140.33±	2.77±	105.21±
Group 2 After N=6	15.74	22.76	0.29	4.21	6.59	0.09	0.34	0.29	0.42	40.96	0.10	0.10	3.07*	0.11*	2.09*

*Difference significant at P≤0.05.

forestomach impaction. The present observations were in agreement with the finding of Ward *et al.* (1993) who reported lower plasma potassium concentration after treatment with 7.2% saline solution as compared to 0.9% saline solution. Following hypertonic saline solution decreased potassium concentration was reported in sheep (Smith *et al.*, 1985; Walsh and Kramer, 1991) in dogs (Constable, 1994) and in buffaloes (Saxena, 2003).

The present study concludes that isotonic (0.9%) saline solution along with supportive medical treatment has more therapeutic efficacy than hypertonic saline solution in management of forestomach impaction in buffaloes.

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